

In the Claims

1-76 (canceled).

77 (new). A method of expressing a gene of interest in a Chinese Hamster Ovary (CHO) cell comprising culturing a CHO cell comprising a vector under conditions that allow for the expression of said gene of interest, said vector comprising at least one gene of interest and one or more chromatin insulators consisting of SEQ ID NO: 1.

78 (new). The method according to claim 77, wherein said vector further comprises at least one DNA element selected from:

- a) an enhancer, or a functional expression enhancing fragment thereof;
- b) a promoter domain or a functional expression promoting fragment thereof; or
- c) a DNA sequence coding for one or more polypeptides of interest.

79 (new). The method according to claim 77, wherein said vector further comprises one or more DNA sequences coding for regulatory elements selected from 5'UTRs, introns, 3'UTRs, mRNA 3' end processing sequences, polyadenylation sites, and internal ribosome entry sequences (IRES).

80 (new). The method according to claim 78, wherein the DNA sequence is coding for more than one polypeptide of interest through a polycistronic mRNA.

81 (new). The method according to claim 77, wherein said vector further comprises one or more DNA elements selected from boundary elements, locus control regions (LCRs), matrix attachment regions (MARs), and elements for recombination and cassette exchange.

82 (new). The method according to claim 78, wherein the promoter is selected from cellular or viral/phage promoters such as mCMV-IE1, mCMV-IE2, hCMV, SV40, RSV, T7, T3, or a functional expression promoting fragment thereof.

83 (new). The method according to claim 78, wherein the polypeptide of interest is selected from FSH, LH, CG, TSH, growth hormone, interferon, TNF binding protein I, TNF binding protein II, IL-18BP, IL-6, IFNAR1, LIF or muteins, fragments, functional derivatives, fusion proteins thereof.

84 (new). The method according to claim 78, wherein the polypeptide of interest is selected from EPO, G-CSF, GM-CSF, a chain of a humanized antibody, a cytokine, a coagulation factor, etanercept, tPA, an integrin or muteins, fragments, functional derivatives, fusion proteins thereof.

85 (new). The method according to claim 78, wherein the polypeptide of interest is selected from adenosine deaminase (ADA), aminoglycoside phosphotransferase (neo), dihydrofolate reductase (DHFR), hygromycin-B-phosphotransferase (HPH), thymidine kinase (tk), xanthine-guanine phosphoribosyltransferase (gpt), multiple drug resistance gene (MDR), ornithine decarboxylase (ODC) and N-(phosphonacetyl)-L-aspartate resistance (CAD), puromycin acetyltransferase (PAC), galactokinase, human folate receptor, or reduced folate carriers.

86 (new). The method according to claim 78, wherein the polypeptide of interest is selected from luciferase, green fluorescent protein, alkaline phosphatase, and horseradish peroxidase or combinations thereof.

87 (new). The method according to claim 78, wherein one insulator is positioned upstream and one insulator is positioned downstream of the DNA sequence coding for a polypeptide of interest.

88 (new). The method according to claim 78, wherein at least two insulators are positioned upstream and downstream of a DNA sequence coding for a polypeptide of interest, respectively.

89 (new). The method according to claim 78, wherein at least two DNA coding sequences are positioned between the insulators.

90 (new). The method according to claim 78, wherein the at least two coding sequences code for subunits of a multimeric protein.

91 (new). The method according to claim 90, wherein the multimeric protein is a hormone comprising a first subunit that is the alpha chain and a second subunit that is the beta chain of a hormone selected from human FSH, human LH, human TSH and human CG.

92 (new). The method according to claim 90, wherein the multimeric protein is an antibody comprising a first subunit that is the heavy chain of an immunoglobulin and a second subunit that is the light chain of an immunoglobulin.

93 (new). The method according to claim 77, wherein said CHO cell simultaneously expresses of two or more genes of interest.

94 (new). The method according to claim 77, further comprising the step of isolating the polypeptide of interest from the CHO cells.

95 (new). The method according to claim 83, wherein said protein of interest is IL-18BP.